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Effect of Dietary Monensin Supplementation on Methane and Nitrous Oxide Emissions from Fresh Dung of Non Pregnant Non Lactating Dry Buffaloes

Shiva Gupta[1*], Madhu Mohini[2], S. S. Thakur[2] and Goutam Mondal[2]

[1]Veterinary Assistant Surgeon, Madhya Pradesh, India
[2]Division of Animal Nutrition, National Dairy Research Institute, Karnal – 132001, Haryana, India

Abstract

The present study was conducted to evaluate the effect of dietary monensin supplementation on methane and nitrous oxide emissions from fresh dung of non pregnant non lactating dry buffaloes. Fourteen dry Murrah buffaloes were randomly divided into two groups of seven animals each based on body weight. Both groups were fed as per ICAR without and with monensin supplementation (350 mg/head/day) in control and treatment group, respectively for sixty days. The daily dry matter intake (kg/d) was similar (P>0.05) in both the groups. Methane emission (g/kg DM, g/kg OM and g/kg NDF) from anaerobic incubation of fresh dung (24 h at 39°C) was decreased in treatment group 0.36, 0.45, 0.58 as compared to control 0.45, 0.56, 0.73, but difference was not significant. N_2O production (mg/kg DM, mg/kg OM and mg/kg N) from fresh dung were lower for treatment group 0.02, 0.11 and 4.99 than control 0.016, 0.13 and 5.56 but difference was not statistically significant. In conclusion, dietary monensin supplementation to non pregnant dry Murrah buffaloes decreased (P>0.05) methane and nitrous oxide emission from fresh dung which will reduce the contribution of buffaloes to nitrous oxide emissions and its negative impact on environment.

Keywords: Dung, Methane, Monensin, Nitrous Oxide

1. Introduction

Livestock are important source of anthropogenic methane (CH_4) emissions at global level. Both, enteric fermentation and manure CH_4 together contributes 80% of agricultural CH_4 emission and about 35-40% of total anthropogenic CH_4 emissions[20]. In ruminant production systems, enteric CH_4 emission is the largest contributor to GHG emissions followed by CH_4 emissions from manure. Methane emissions from animal manure depend on its consistency (solid, liquid) and management (collection, storage, spreading). Steinfeld et al. reported that Globally Greenhouse Gas (GHG) emissions from manure management to be 2.2 giga ton of CO_2 equivalents per year. In India, 127 Giga gram methane emissions from manure storage were estimated (INCAA). Methane emissions from manure occur under anaerobic conditions and less CH_4 is produced when manure handled as a solid or when deposited on pasture or rangelands (USEPA). Organic matter present in manure forms Volatile Fatty Acids (VFAs) by hydrolysis and on storage methanogens results in the formation of methane. Nielfa et al. reported that methane production increased with the increase in the organic matter (volatile solids) content

*Author for correspondence

of the manure. Moller et al. reported that higher CH_4 produced from pig (356 L/kg VS) and sow (275 L/kg VS) manure compared to dairy cattle manure (148 L/kg VS). In anaerobic conditions i.e. liquid system of storage more CH_4 production occurs while in solid system CH_4 emission is lesser because of aerobic condition but during rainy season CH_4 emission from the solid system may increase due to increase in moisture as well.

Nitrous oxide (N_2O) is produced from the combined nitrification-denitrification process that occurs on the Nitrogen (N) in manure. The majority of N in manure is in ammonia (NH_3) form. Nitrification occurs aerobically and converts this ammonia into nitrate, while denitrification occurs anaerobically and converts the nitrate to nitrous oxide. Temperature, pH, biochemical oxygen demand and nitrogen concentration affects N_2O generation. Erickson and Klopfenstein reported lower nitrogen excretion and lower nitrogen volatilization losses of phase-fed cattle. Biochemical Oxygen Demand (BOD) is the amount of dissolved oxygen used by aerobic microorganisms to completely consume the available organic matter. Increasing aeration initiates the nitrification-denitrification reactions, and hence makes release of nitrous oxide possible and N_2O production is expected to increase with increasing aeration. Rahman et al. reported N_2O emission rates 26 g/head/day from the pen surface of feedlot beef cattle and somewhat lower N_2O emissions (0.01 kg/cow/d) were reported for an open-lot dairy in southern Idaho by Leytem et al. Use of high protein grass increased N_2O losses in farmyard manure particularly at the start of storage from grass fed cows when the hay rations[9].

Boadi et al. observed no significant difference in manure pack N_2O emissions from low forage: Grain and high forage: grain diets in feedlot cattle. Kulling et al. reported 0.06 to 1.49 g/cow/d nitrous oxide emissions during seven weeks storage from slurry and farmyard manure. Harrison et al. examined that grazing of Leucaena increased N_2O emissions by more than 38% however; CH_4 emissions were reduced at whole farm level. CH_4 and N_2O production and emissions from manure depends on feed composition, its digestibility, animal species and their physiology, manure management practices and meteorological conditions like temperature, precipitation, wind, etc. (Brown et al.). IPCC reported default values for methane EF as 5 kg/head/year for cattle and buffalo for manure irrespective of their age and productivity. Gupta et al. reported average CH_4 and N_2O emissions were 6.6 mg/kg dung/day and 35.42 µg/kg dung/day from solid storage (as a heap) of bovine manure from different seasons, during field measurements in India. They also calculated CH_4 EF (kg/head/year) was 1.2, 2.3, 2.7 and 3.3 for 0-1 year, 1-3 year, adult and dairy buffalo, respectively. They also calculated EFs for N_2O (mg/head/year) which were 5.2, 9.9, 11.7, 11.0 for 0-1 year, 1-3 year, adult and dairy buffalo, respectively in India for year 2000. They also reported CH_4 and N_2O EFs from solid storage of manure varied from 0.8 to 3.3 kg CH_4/head/day and 3 to 11.7 mg N_2O/head/year for bovine and these values were lower than IPCC default values. NATCOM reported CH_4 EF for buffalo manure management were 1.8, 3.4, 4 and 4.4 kg/year for 0-1 year, 1-3 year, adult and dairy buffalo, respectively in India. Only a few studies have been carried out on efficacy of monensin supplementation on methane and nitrous oxide emissions from fresh dung of non pregnant non lactating dry buffaloes.

2. Material and Methods

The experimental protocol was approved by Institutional Animal Ethics Committee (IAEC/03/16 dated 19.05.2016) of the National Dairy Research Institute, Karnal, India. The maximum ambient temperature goes up to 45°C during summer, minimum about 5°C during winter, relative humidity varies from 18 to 97% percent with an annual rain fall is approximately 760-960 mm most of which is received during the months of July to August. (Central Soil Salinity Research Institute, Karnal, Haryana). The present experiment was conducted during mid-june to mid-august. Fourteen dry Murrah buffaloes having average body weight of 597.62 kg were selected from the Institute Livestock Research Centre and identified by numbered ear tags, tethered with nylon rope individually in a well-ventilated stall (floor space = 4 m^2per animal) provided with uniform management practices and having facilities for individual feeding. Animals were dewormed using Fenbendazole (Panacur, Intervet, India) at 10 mg/kg BW and treated against ectoparasites using Deltamethrin (Butox) spray 10 d before the commencement of experimental feeding. After an adaptation period of 10 days, animals were randomly divided into two groups of seven animals in each on the basis of body weight. Both groups were fed ration comprising of green sorghum fodder chopped at 2–3 cm length, concentrate mixture (in g/kg as mixed: maize 330, groundnut cake 180, mustard oil cake 100, cotton seed cake 50, wheat bran

200, de-oiled rice bran 60, bajra 50, mineral mixture 20 and common salt 10) and wheat straw, to meet their nutrient requirements according to ICAR without and with monensin supplementation (350 mg/head/day) in control and treatment group, respectively for sixty days. Monensin was top dressed on concentrate mixture in the form of Rumensin (Elanco, Division of Eli Lilly and company (NZ Limited), which contains monensin in a concentration of 20% Mill mix (Equivalent to 200 g of monensin activity as monensin sodium per kg). All animals were provided clean and fresh drinking water twice daily in morning at 10.00 h and evening at 17:30 h. After 55 days of experimental feeding trial, during which daily output of fresh feces were collected from each group and mixed thoroughly and sampled once daily 1% of thoroughly mixed total fecal matter by fresh weight were used for Dry Matter (DM) determination by drying at 60°C for 48 h to constant weight. Then, dried fecal sample used for organic matter analysis. For nitrogen (N) determination (Kjeldahl method) feces samples (1/500 of daily voidance) were preserved in 30% sulphuric acid to make pooled samples of 7 d for individual animals.

Fresh dung methane and nitrous oxide emissions were estimated (Nampoothiri). Fresh dung was collected from the composite dung of the animals in each group, in bottles of 2 litre capacity and was sealed airtight with the help of rubber cork and aluminium cap. It was done in 3 replicates daily for seven days. Bottles were kept in an incubator maintained at 39°C. Total gas production and sample was collected after 24 hrs for estimation of methane and nitrous oxide with the help of 3 way cork and 100 mL calibrated glass syringes (Hamilton syringes). Measurement of methane and nitrous oxide was done with gas liquid chromatography, using FDI detector for methane and ECD detector for nitrous oxide. To determine the methane concentration, collected gas sample in the syringes by piercing the silicon tube fitted with the in vitro syringes and 5 mL of gas sample was immediately injected into the gas chromatograph (Nucon 5700, Nucon Engineers, New Delhi) instrument equipped with a flame-ionization detector (100°C) and stainless steel column packed with Porapak-Q (length 1.5; o.d. 3.2 mm; i.d. 2 mm; mesh range 80-100). The column and injector temperatures were 50 and 40°C in both the instruments. All samples were analyzed in duplicate except standards, which were analyzed in triplicate. Nitrogen was used as the carrier gas at a pressure of 1 kg/cm^2. The standards were run in the beginning and end of each day with the methane standard run every 10 samples throughout the day.

Gas concentrations (N_2O and CH_4) were determined from peak areas and identified from their different retention times relative to the known standards. The methane output calculated using following formula:

Conc. of methane (ppm) in sample =

$$\frac{\text{Area of methane in sample}}{\text{Area of methane in standard}} \times \text{Conc.of methane in standard}$$

N_2O was analyzed by gas chromatography (Nucon 5700, Nucon Engineers, New Delhi), fitted with an electron capture detector to determine N_2O with 3.3 m molecular sieve column with an i.d. of 0.32 mm. The column and injector temptratures were 50 and 40°C and temprature of detector was 350°C. All samples were analyzed in duplicate except standards, which were analyzed in triplicate. Nitrogen was used as the carrier gas at a pressure of 1kg/cm^2. Gas concentrations were determined from peak area and identified from their different retention times relative to the known standards.

Conc. of nitrous oxide (ppm) in sample =

$$\frac{\text{Area of nitrous oxide in sample}}{\text{Area of nitrous oxide in standard}} \times \text{Conc.of nitrous oxide in standard}$$

Data collected during study were subjected to the statistical analysis as per Snedecor and Cochran. Independent sample t-test was done to find out the significant difference between groups using software package IBM SPSS statistics version 16.0, 2010.

3. Results and Discussion

Chemical composition of ingredients of basal diet is presented in Table 1. The chemical composition of all the ingredients were within normal range reported previously (Das et al., Prusty and Sharma).

Dry matter, organic matter and nitrogen contents of fresh dung from both groups are presented in Table 2. The organic matter content and nitrogen content were lower in treatment group as compared to control.

Data pertaining to daily dry matter intake, methane and nitrous oxide emissions from fresh dung of non pregnant dry buffaloes are presented in Table 3. Daily dry matter intake was similar (P>0.05) in both the groups. Methane emission (g/kg dry matter) from anaerobic incubation of fresh dung (24 h at 39°C) was decreased in monensin supplemented

group (0.36) as compared to control (0.45) but difference was not significant. Methane emission g/kg organic matter and g/kg neutral detergent fiber from dung were also lower in monensin supplemented group as compared to control this might be due to lower organic matter content of fresh dung of treatment group as compared to control. Methane production increased with the increase in the organic matter (volatile solids) content of the manure (Nielfa et al.,

Table 1. Chemical composition and energy contents of offered feedstuffs

Parameter (%)	Concentrate Mixture	Wheat straw	Sorghum Green fodder
DM	90.39	90.63	28.08
OM	94.54	89.87	89.25
CP	21.78	3.70	10.09
EE	3.9	1.30	1.93
TA	5.45	10.13	10.75
NDF	24.1	77.11	64.38
ADF	11.49	51.32	34.22
TDN	76.33	43.61	52.66
DE (MJ/kg DM)	14.08	8.05	9.71
ME (MJ/kg DM)	12.34	6.24	7.93

Table 2. Dry matter, organic matter and nitrogen contents of fresh dung from both groups

Dung parameters (%)	Control	Treatment
Moisture	81.25 ± 0.60	81.88 ± 0.48
Dry Matter	18.75 ± 0.60	18.12 ± 0.48
Organic Matter	81.22 ± 0.35	81.00 ± 0.28
Nitrogen	1.91 ± 0.05	1.90 ± 0.10
Total Ash	18.78 ± 0.35	19.00 ± 0.28

Table 3. Effect of Monensin supplementation on nutrient intake, methane and nitrous oxide emissions from fresh dung of non pregnant dry buffaloes

Parameter	Control	Treatment	P value
DMI (kg/d)	9.60 ± 0.08	9.24 ± 0.16	0.06
CPI (kg/d)	0.70 ± 0.01	0.66 ± 0.03	0.18
TDNI (kg/d)	5.46 ± 0.12	5.08 ± 0.24	0.19
Total gas (L/kg DM)	2.96 ± 0.04	2.43 ± 0.03	0.34
Total gas (L/kg OM)	3.64 ± 0.05	2.99 ± 0.04	0.33
CH4 g/kg Dung	0.086 ± 0.01	0.066 ± 0.01	0.42
CH4 (g/kg Dung DM)	0.45 ± 0.07	0.36 ± 0.11	0.53
CH4 (g/kg Dung OM)	0.56 ± 0.09	0.45 ± 0.13	0.52
CH4 (g/kg Dung NDF)	0.73 ± 0.12	0.58 ± 0.18	0.52
N2O (mg/kg Dung DM)	0.020 ± 0.006	0.016 ± 0.004	0.6
N2O (mg/kg Dung DM)	0.11 ± 0.03	0.09 ± 0.02	0.68
N2O (mg/kg Dung OM)	0.13 ± 0.04	0.11 ± 0.03	0.68
N2O (mg/kg Dung N)	5.56 ± 1.50	4.99 ± 1.33	0.78

2015). Nitrous oxide (N_2O) production (mg/kg dry matter, mg/kg organic matter and mg/kg nitrogen) from fresh dung were lower (P>0.05) for monensin supplemented group 0.02, 0.11 and 4.99 than control 0.016, 0.13 and 5.56, this might be due to lower nitrogen content of fresh dung of treatment group as compared to control. Erickson and Klopfenstein reported lower nitrogen excretion and lower nitrogen volatilization losses of phase-fed cattle which support present study findings. Nampoothiri reported lower emissions of methane (0.08 g CH_4 /kg dry matter) and higher nitrous oxide emissions (0.04-0.05 N_2O mg/kg dry matter) from fresh dung of growing male buffalo calves fed with different ratios of berseem, wheat straw and concentrate mixture as per requirement, this increase methane emission from dung was attributed to an increase in readily fermentable carbohydrates in dung from high concentrate diet in growing animal as compared to dry buffaloes which were fed as close to maintenance. Boadi et al. also reported that feeding high concentrate lead to high methane emissions which support findings of present study. According to Kulling et al. use of high protein grass increased nitrous oxide losses in farmyard manure particularly at the start of storage from grass fed cows, this support present study findings of lower nitrous oxide emissions as compared to reported by Nampoothiri. Rajvaidya also reported lower CH_4 emissions 0.01 g/kg DM from fresh dung of growing male buffalo calves fed with different ratios of Leucaena leucocephala leaves, WS and concentrate mixture. This difference might be due to less amount of concentrate fed close to maintenance in dry buffaloes in comparison to other studies on growing calves.

4. Conclusions

Methane emission (g/kg dry matter) and nitrous oxide (N_2O) production (mg/kg dry matter) from anaerobic incubation of fresh dung (24 h at 39°C) was non significantly decreased in monensin supplemented group as compared to control. The number of animals per treatment was very small, and more long term studies are needed for evaluating efficacy of monensin on methane and nitrous oxide mitigation from fresh dung in non pregnant dry Murrah buffaloes.

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